FINAL REPORT

Microbiological Sampling Report

for

National Oceanic & Atmospheric Administration

Samplings Conducted on the Twelfth Floor
of Building SSMC-3
on February 17, 2000

Interagency Agreement #: D8H00CO31200
Task: 9903

January 16, 2001

Prepared by
US Public Health Service
Division of Federal Occupational Health

Bethesda Central Office

Executive Summary

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 12242 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from these rooms and an indoor reference room 12302. Air samples were also collected from outdoors.

Findings are as follows:

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, contact plate, and dust samples.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Very low fungal burden was detected from swab samples collected from surfaces of supply diffusers and return troughers in light fixture.

· Fungal levels in plenum dust of these rooms were at 10^3 CFU/g of fine dust levels. *Penicillium* dominated these samples.

· The fungal level in furniture dust of room 12242 was at 10^3 CFU/g level. *Cladosporium* was the predominant fungal genus detected.

INTRODUCTION

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 12242 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from these rooms and an indoor reference room 12302. Air samples were also collected from outdoors.

EVALUATION METHODOLOGY

Air Samples

Various types of samples were collected from these rooms on February 17, 2000. Two types of air samples were collected from each room: (1) culturable method using Andersenâ N-6 samplers at a flow rate of 28.3 L/min, and (2) non-culturable method using Zefonâ Air-O-Cell cassettes at a flow rate of 15 L/min. Indoor Andersenâ air samples were collected for 3 minutes and outdoor samples were collected for both one and three minutes. Two percent (2 %) malt extract agar (MEA) and cellulose Czapek agar (CCA) was used to recover general fungi and cellulose-loving fungi, respectively. Non-culturable air samples were collected at the aforementioned sampling locations. Indoor samples were collected for ten minutes and outdoor samples were collected for both five and ten minutes. Outdoor air samples were collected near the entrance of the building. Temperature and relative humidity measurements were collected from each air sampling location by a battery operated, direct readout Hygroskopâ meter.

Contact Plate Samples

To determine fungal burden on horizontal and vertical surfaces of these rooms, five to eight contact plate samples were collected from each room. Samples were collected from randomly selected horizontal and vertical surfaces. Sampling was conducted by pressing the MEA-filled Rodacâ plate against the surface of interest for five seconds. A total of 13 contact plate samples were collected.

Swab Samples

Swab samples were collected from surfaces of each supply diffusers and return troughers in each room. They were collected by wiping a known area of surface with a sterile cotton swab (Culturetteâ) wetted with holding media. Approximately 5 in^2 area was wiped for return trougher and 4 in^2 for supply diffusers. The swab was then placed directly into its holder. Each holder was labeled with an identifiable number. A total of 14 wipe samples were collected from these rooms.

Vacuum Dust Samples

Dust accumulated on chairs and fabric system furniture, and the plenum were collected with a High Efficiency Particulate Air (HEPA) vacuum attached with a special “sock” device. Total surface areas of 9 ft^2 were vacuumed from system furniture and chairs, and composite as one sample. Dust accumulated above the ceiling plenum was also vacuumed and composite as one sample. Vacuum time was at least five minutes. One composite plenum sample was collected from each room. A composite furniture sample was collected from room 12242.

All samples collected were sent for next morning delivery to FOH's Environmental Microbiology Laboratory (EML) in Philadelphia, Pennsylvania for analysis.

Laboratory Procedures

Upon receipt, all Andersenâ air and contact plate samples were incubated in a 25˚C incubator. Each swab sample was suspended in sterile distilled water, diluted serially, and inoculated onto agar plates. Both MEA and CCA were used for retrieving fungi. At least three dilution series were used for each sample. Each vacuum dust sample was sieved through a 250 mm sieve. The fine dust (< 250 mm) retrieved was then weighed and followed the dilution plating for fungal analysis.

All plates were incubated in a 25˚C incubator. They were examined every other day for up to 10 days to ensure the full recovery of fungi. Fungal identification was based on colony morphology, spores and conidia formation. Total fungal colonies formed on each MEA plate and *Stachybotrys chartarum* on CCA plates were counted and recorded. Fungal levels in samples were presented as colony forming units (CFUs) per measuring unit. For example, CFU/m^3 for Andersenâ air samples, CFU/in^2 for wipe samples, CFU/plate for contact plate samples, and CFU/g of fine dust for vacuum dust samples.

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All Zefonâ cassette samples were analyzed by the Environmental Microbiology Laboratory in Escondido, California for direct microscopic examination. Fungal spores were identified and their airborne levels were presented as spores/m^3.

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RESULTS AND DISCUSSION

Temperature and Relative Humidity

Indoor temperature and relative humidity measurements ranged from 72.3°F to 72.6°F, and 21.0% – 28.6%, respectively (Table 1). Outdoors temperature reading was lower (47.5°F). Outdoor relative humidity was 20.9 % (Table 1).

Microbiological Analyses Results

All laboratory analytical reports from FOH's EML are presented in Attachment A in a laboratory report #NOAA-00-34R. Results from microscopic examination of Zefonâ cassette samples are presented in Attachment B.

Air Samples

Andersen Results

Outdoor airborne fungal levels were higher than those of indoors (Table 1). *Cladosporium* dominated outdoor fungal flora. Other fungi detected were *Penicillium, Epicoccum, Paecilomyces,* and Basidiomycetes. Fungi recovered indoors were Basidiomycetes. *Stachybotrys chartarum* was not detected from these samples.

Zefon Results

Outdoor airborne fungal spore levels were higher than those of indoors (Table 1). Fungal spore types detected from outdoors were *Cladosporium* and Basidiospores. Fungal spores detected indoors were rust, smut, and *Pithomyces. Stachybotrys chartarum* was not detected from any sample collected.

Table 1. Temperature and relative humidity measurements and airborne fungal levels at different rooms of the 12th floor in SSMC-3 on February 17, 2000.

Rooms **Parameters**	**12302** **Reference#**	**12242**	**Outdoors**
Temperature (°F)	72.6	72.3	47.5
Relative Humidity (%)	21.0	28.6	20.9
Airborne Fungal Levels (CFU/m^3)	12	<12	200* 212
Total Fungal Spores (Spores/m^3)	20	14	294* 280

Indoor reference.

* Two samples were collected from outdoors.

Swab Samples

Fungal levels on swab samples were very low. Most (13 out of 14) samples collected from surfaces of

supply diffusers and return troughers in light fixtures were below the detection limits (BDL) (2 CFU/in^2 for supply diffuser and 3 CFU/in^2 for return trougher). The sample showed fungal growth (3 CFU/in^2) was collected from room 12242 on supply diffuser.

Contact Plate Samples

In general, higher fungal levels were detected from the horizontal surfaces than vertical surfaces (Table 2). Fungal levels on vertical surfaces ranged from BDL of 1 CFU/plate to 1 CFU/plate. Fungal levels on horizontal surfaces ranged from 5CFU/plate to 12 CFU/plate. Fungi recovered from these samples were *Penicillium, Cladosporium, Aspergillus, Alternaria, Aureobasidium, Pithomyces,* and Basidiomycetes. *Stachybotrys chartarum* was not detected from these samples.

Vacuum Dust Samples

Fungal levels in the fine dust collected from these rooms were at 10^3 CFU/g of fine dust levels (Table 3). *Stachybotrys chartarum* was not detected from these samples (Table 3).

Table 2. Fungal levels (CFU/plate) on horizontal and vertical surfaces of different rooms at the 12th floor of SSMC-3, by contact plate sampling collected on February 17, 2000.

Rooms	**12302** **Reference#**	**12242**
Horizontal Surfaces (CFU/plate)	4 – 12* (4**)	5 (1)
Vertical Surfaces **(CFU/plate)**	<1 - 1 (4)	<1 (4)

Indoor reference.

* Ranges.

** Total sample number.

Plenum Dust

Penicillium was the predominant fungal genera detected from these samples. Other fungi detected, in a descending order, were *Aspergillus niger, Alternaria*, *Paecilomyces*, *Cladosporium*, and *Rhizopus.*

Furniture Dust

Predominant fungal genus detected from furniture dust collected from room 12242 was *Cladosporium.* Other fungi recovered were *Trichoderma*, *Alternaria*, and *Aspergillus niger*, and *Epicoccum*

Table 3. Total fungal levels (CFU/g of fine dust) in fine dust collected from carpet, plenum, and furniture of rooms 12242 and 12302 of SSMC-3, by vacuum dust sampling, collected on February 17, 2000.

Rooms	**12302** **Reference#**	**12242**
Plenum **(CFU/g of fine dust)**	2,000 (-*)	5,941 (-)
Carpet **(CFU/g of fine dust)**	NA**	NA
Furniture **(CFU/g of fine dust)**	NA	3,200 (-)

Indoor reference.

*+: *Stachybotrys chartarum* was detected on MEA and/or CCA plates.

-: *Stachybotrys chartarum* was not detected on MEA and CCA plates.

** Not applicable.

CONCLUSIONS

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, contact plate, and dust samples.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Very low fungal burden was detected from swab samples collected from surfaces of supply diffusers and return troughers in light fixture.

· Fungal levels in plenum dust of these rooms were at 10^3 CFU/g of fine dust levels. *Penicillium* dominated these samples.

· The fungal level in furniture dust of room 12242 was at 10^3 CFU/g level. *Cladosporium* was the predominant fungal genus detected.

RECOMMENDATIONS

· Conduct a thorough cleaning of these rooms by HEPA vacuuming and wet wiping.

· Conduct any above ceiling plenum work after hour. Thoroughly HEPA vacuum the surrounding areas afterwards.

· Implement an emergency water intrusion protocol for this building to adequately manage any unexpected water intrusion in order to prevent fungal proliferation.

ATTACHMENT A

Microbiological laboratory report #NOAA-00-34R for samples collected

from twelfth floor of SSMC-3, on February 17, 2000.

ATTACHMENT B

Results from microscopic examination of Zefon air samples collected

from twelfth floor of SSMC-3, on February 17, 2000.

USPHS DFOH ENVIRONMENTAL MICROBIOLOGY LABORATORY, PHILADELPHIA, PA

LABORATORY REPORT #NOAA-00-34R

Client agency: National Oceanic and Atmospheric Administration, Silver Spring, MD

POIS#/task #: D8H00CO31200 / 9903

Sampling date: 2/17/00

Dates of inoculation: 2/17/00 (airs and contact plates), 2/18/00 (wipes), and 2/20/00 (dust)

General location: SSMC-3, Silver Spring, MD

Specific location: 12th floor

Sampling techniques: Air (Andersen N-6 sampler), contact plate, wipe, and vacuum dust samplings

Medium used: Malt extract agar (MEA) and cellulose Czapek agar (CCA) for fungi

Samples submitted by: J. Sobelman

Date characterization completed: 3/1/00

(A) Air samples on MEA and CCA plates

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
3-12302-0217A1, A2	12th floor, room 12302, center of office	84.9	1. Basidiomycetes (1*) CFU/m^3 = 12	No
3-12242-0217A1, A2	12th floor, room 12242, center of room	84.9	No fungal growth CFU/m^3 < 12	No

3-OA1-0217, 3-OA2-0217	Outside bldg. 3	84.9	1. *Cladosporium* (13) 2. *Penicillium* (2) 3. *Epicoccum* (1) 4. *Paecilomyces* (1) CFU/m^3 = 200	No
3-OA3-0217, 3-OA4-0217	Outside bldg. 3	28.3	1. *Cladosporium* (5) 2. Basidiomycetes (1) CFU/m^3 = 212	No
FB	Field blank	NA#	No fungal growth	No
SB	Shipping blank	NA	No fungal growth	No

(B) Contact plate samples on MEA plates

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
3-12302-0217CP1	12th floor, room 12302, S wall	1. *Rhizopus* (1) **CFU/plate = 1**
3-12302-0217CP2	12th floor, room 12302, N wall	No fungal growth CFU/plate < 1
3-12302-0217CP3	12th floor, room 12302, E wall	No fungal growth CFU/plate < 1
3-12302-0217CP4	12th floor, room 12302, W wall	1. *Penicillium* (1) **CFU/plate = 1**
3-12302-0217CP5	12th floor, room 12302, top of desk	1. *Penicillium* (6) 2. *Alternaria* (2) 3. *Aspergillus sp.* (2) 4. *Cladosporium* (2) **CFU/plate = 12**

3-12302-0217CP6	12th floor, room 12302, top of slide case	1. *Penicillium* (2) 2. *Alternaria* (1) 3. *Cladosporium* (1) **CFU/plate = 4**
3-12302-0217CP7	12th floor, room 12302, top of book case	1. *Penicillium* (3) 2. *Alternaria* (1) 3. *Paecilomyces* (1) **CFU/plate = 5**
3-12302-0217CP8	12th floor, room 12302, floor	1. *Cladosporium* (4) 2. *Penicillium* (3) 3. *Aureobasidium* (1) 4. *Pithomyces* (1) 5. Basidiomycetes (1) **CFU/plate = 10**

Sample ID	**Sampling Location**	**Fungi detected on MEA @ 25º C**
3-12242-0217CP1	12th floor, room 12242, S wall	No fungal growth CFU/plate < 1
3-12242-0217CP2	12th floor, room 12242, N wall	No fungal growth CFU/plate < 1
3-12242-0217CP3	12th floor, room 12242, E wall	No fungal growth CFU/plate < 1
3-12242-0217CP4	12th floor, room 12242, W wall	No fungal growth CFU/plate < 1
3-12242-0217CP5	12th floor, room 12242, floor tile	1. *Aspergillus niger*** (2) 2. *Alternaria* (1) 3. *Aureobasidium* (1) 4. *Penicillium* (1) **CFU/plate = 5**

(C) Wipe samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
Blank	Blank	Blank	NA	10X-MEA 10X-CCA	No fungal growth	No
W37	3-12302-0217R1	12th floor, room 12302, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W38	3-12302-0217R2	12th floor, room 12302, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W39	3-12302-0217S1	12th floor, room 12302, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
W40	3-12242-0217R1	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W41	3-12242-0217R2	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W42	3-12242-0217R3	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W43	3-12242-0217R4	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W44	3-12242-0217R5	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W45	3-12242-0217R6	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No

W46	3-12242-0217R7	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W47	3-12242-0217R8	12th floor, room 12242, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W48	3-12242-0217S1	12th floor, room 12242, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W49	3-12242-0217S2	12th floor, room 12242, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W50	3-12242-0217S3	12th floor, room 12242, supply	4	10X-MEA 10X-CCA	1. *Penicillium* (1) CFU/in^2 = 3	No

(D) Vacuum dust samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
V10	3-12302-0217V01	12th floor, room 12302, above ceiling	0.100	40X-MEA 10X-CCA	**1. *Penicillium* (4)** 2. *Paecilomyces* (1) **CFU/g = 2,000**	No
V11	3-12302-0217V02	12th floor, room 12302, furniture	0.100##	40X-MEA 10X-CCA	**1. *Cladosporium* (9)** 2. *Trichoderma* (3) 3. *Alternaria* (2) 4. *Aspergillus niger*** (1) 5. *Epicoccum* (1) **CFU/g = 3,200**	No

V12	3-12242-0217V01	12th floor, room 12242, above ceiling	0.101	40X-MEA 10X-CCA	**1. *Penicillium* (8)** 2. *Aspergillus niger*** (3) 3. *Alternaria* (2) 4. *Cladosporium* (1) 5. *Rhizopus* (1) **CFU/g = 5,941**	No

* Colony counts.

** Opportunistic fungi.

*** Toxigenic fungi.

Not applicable.

5ml of sterilized distilled water were added instead of 10ml.